

Earlier bites by uninfected mosquitoes boost West Nile deaths in lab mice

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There's one more reason to try to avoid being bitten by mosquitoes, scientists have discovered: bites from mosquitoes that aren't infected by the West Nile virus may make the disease worse in people who acquire it later from West Nile-infected mosquitoes.

Researchers from the University of Texas Medical Branch at Galveston (UTMB) announced their discovery in a paper published online by the journal PLoS ONE. In the paper, they describe experiments showing that lab mice on which mosquitoes have previously fed are far more likely to die from West Nile infection than are mice unexposed to such mosquito bites.

The effect is induced by mosquito saliva, according to UTMB professor Stephen Higgs, one of the paper's senior authors.

"This virus is transmitted from mosquitoes in saliva, and we'd already demonstrated that mosquito saliva has an effect on the vertebrate immune system that makes West Nile infection worse," Higgs said.

"What this new work shows is that the saliva delivered by even earlier feedings can also alter the course of the infection. This is important, because in natural situations in many parts of the world – Southeast Texas, for example — animals and some people are being exposed to mosquito feeding almost continuously."

In their experiments, researchers exposed sedated mice to feeding by between 15 and 20 *Aedes aegypti* mosquitoes for an hour once a week.

Scientists then allowed a single West Nile virus-infected mosquito to feed once on each of these mice and also on each of a control group of mice that were previously unbitten by mosquitoes.

The results were striking: 68 percent of mice exposed to two weekly mosquito feedings died of West Nile virus, and those exposed to four weekly mosquito feedings suffered a 91 percent mortality rate. By contrast, the virus killed only 27 percent of the mice previously unexposed to saliva from mosquitoes that were free of West Nile infection. Analyses of responses of the mouse immune systems also showed a strong contrast between the previously exposed and unexposed mice.

“When we examined the immune reactions, one that stood out was an increase in the immune signaling molecule interleukin-10,” said Brad Schneider, the paper’s lead author and a UTMB alumnus who is now a postdoctoral fellow at the Institut Pasteur in Paris. “This host response to the saliva of the mosquito causes a shift in the immune response at the site where the virus first contacts the host, and the virus takes advantage of this shift.”

The UTMB researchers were surprised to find that mosquito bites seemed to have a detrimental effect with West Nile virus, because multiple earlier bites from other uninfected arthropods can actually protect against the parasites and bacteria carried by them. “Previous work has clearly indicated that pre-exposure to the bites of uninfected sand flies has a protective effect for mice against cutaneous leishmaniasis,” said Dr. Lynn Soong, the paper’s other senior author and an immunologist who works on the sand fly-transmitted protozoan parasite infection, dubbed “Baghdad boil” by American troops in the Middle East.

“Since this goes against the work we’ve seen with both bacteria and

parasites, we definitely didn't expect this result," Schneider said. "But when we stood back and looked at it, it made sense. For a parasite or bacterium, the influx of immune cells brought in by this inflammatory response would be negative, but with the West Nile virus, you're just giving it more susceptible cells to infect."

Both Higgs and Schneider emphasized that the mouse experiments offered no definitive answers to the question of human responses to West Nile. "This is a mouse model, but that's the best we've got at the moment," Higgs said. "The thing is, it suggests that there may be yet another reason to avoid mosquitoes, to tidy up your yard and wear mosquito repellent."

Source: University of Texas

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